The Biological Use of Platelet-Rich Plasma in Skeletal Muscle Injury and Repair

Jordan Boivin,* BS, MD, Rachael Tolsma,† BS, Peter Awad,† BS, Keith Kenter,* MD, and Yong Li,*‡ MD, PhD
Investigation performed at Western Michigan University Homer Stryker MD Medical School, Kalamazoo, Michigan

Platelet-rich plasma (PRP) is a blood product that contains several growth factors and active proteins. PRP is thought to be used autologously to assist in the repair of injured tissues as well as to treat pain at the site of injury. The mechanism behind PRP in regenerative medicine has been well investigated and includes the identification and concentration of released growth factors and exosomes. The benefits of PRP have been highly recommended and are used widely in orthopaedics and sports medicine, including repair of injured skeletal muscle. This current report summarizes some of the more recent studies in the use of PRP as it relates to muscle healing, in both the in vitro and clinical arenas.

Keywords: biologic healing enhancement; muscle injuries; platelet-rich plasma

THE BIOLOGICS OF PRP

The Conception of PRP

Platelet-rich plasma (PRP) is an autologous blood product that contains supraphysiologic concentrations of platelets and is used in orthopaedics for pain control and tissue regeneration. Two types of PRPs, leukocyte-poor PRP (LP-PRP) and leukocyte-rich PRP (LR-PRP), have been determined.[2] To prepare PRP, blood is drawn from a patient and then processed by 1 of many commercial preparation kits, which typically includes several rounds of centrifugation (Figure 1).[13,47] PRP was originally used by hematologists to treat thrombocytopenia and was then applied in oral-maxillofacial surgery and found to improve bone healing.[3,45,50,62] Because of the positive effect seen on bone, it was investigated for regenerative purposes in orthopaedic surgery and has since been explored in sports medicine, dentistry, cardiac surgery, pediatric surgery, gynecology, urology, plastic surgery, ophthalmology, and dermatology.[3] Contributing to the interest in using PRP for tissue regeneration was a study from Slater et al[58] in the 1990s, which found that human platelet concentrate increased the proliferation of osteoblast-like cells through the release of growth factors. The supraphysiologic concentration of platelets leads to a release of growth factors in normal physiologic proportions, which has since been shown to promote healing by inducing the coagulation cascade, angiogenesis, proliferation and differentiation of resident stem cells, and chemotaxis of macrophages.[15,66] More recently, a study indicated that the platelets facilitate tissue healing with stem cells by mitochondrial transfer and metabolic reprogramming.[33] It is unclear if PRP contains platelets that involve these mitochondrial transfers in tissue healing. In addition to its regenerative potential, PRP is an autologous blood product that has few adverse effects and is generally considered safe.[15] Because of the ease of its preparation and its favorable safety profile, PRP has now been explored as a treatment option for pain relief and the regeneration of many tissue types and pathologies in orthopaedics.

Growth Factors and Platelet Activation

The mechanism by which PRP functions has been heavily researched and is primarily thought to be due to the

‡Address correspondence to Yong Li, MD, PhD, Department of Orthopaedic Surgery, Biomedical Engineering at Western Michigan University Homer Stryker MD School of Medicine, 1000 Oakland Dr, Kalamazoo, MI 49008, USA (email: yong.li@med.wmich.edu).
*Department of Orthopaedic Surgery, Biomedical Engineering at Western Michigan University Homer Stryker MD School of Medicine, Kalamazoo, Michigan, USA.
†Western Michigan University Homer Stryker MD School of Medicine, Kalamazoo, Michigan, USA.
J.B., R.T., and P.A. contributed equally to and share first authorship of the article.
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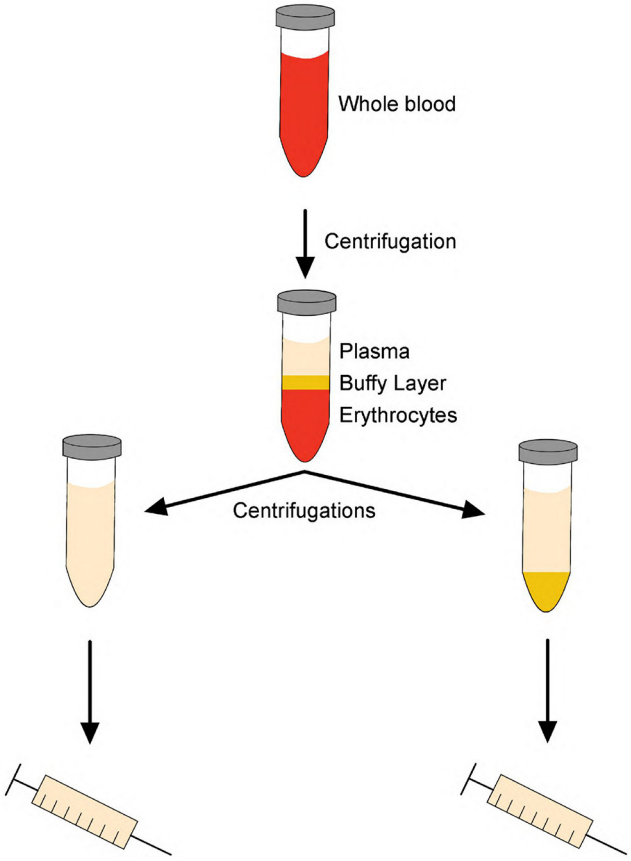

Figure 1. The process of platelet-rich plasma (PRP) preparation. To prepare PRP, the patient's blood is drawn and undergoes serial centrifugation. After the first centrifugation, blood separates into layers: the plasma, buffy layer, and erythrocytes. Leukocyte-rich PRP includes the plasma and the buffy layer, which contains leukocytes, while leukocyte-poor PRP contains only the plasma for the second centrifugation. After processing, these obtained PRPs can be injected back into the patient or applied during orthopaedic surgery.

growth factors released by the contained platelets. These growth factors aid in tissue regeneration by influencing resident cells' migration, proliferation, differentiation, and extracellular matrix (ECM) synthesis.[49] Well-known anabolic growth factors released from the PRP have been reported to include basic fibroblast growth factor (bFGF), insulin-like growth factor 1 (IGF-1), tissue growth factor beta (TGF-β), vascular endothelial growth factor (VEGF), and platelet-derived growth factor (PDGF).[49]

Although growth factors are thought to promote tissue healing, their released concentrations from the PRP decrease over time.[45] Kobayashi et al[31] demonstrated that PRP releases high concentrations of PDGF-AB and TGF-β1 for a short amount of time and therefore may only be useful for the rapid delivery of growth factors. The rapid promotion, release, and decline in growth factors over time necessitates a narrow time window for the delivery of PRP to patients after preparation, which leads to difficulty storing PRP and limitations in clinical application.

Because of the short lifespan of the supraphysiologic concentration of those growth factors, PRP may need an activator added to induce degranulation of the platelets and increase growth factor content.[47] Traditionally, PRP has been activated by calcium gluconate or bovine thrombin.[66] Bovine thrombin presents immunogenicity concerns, which has led to a search for an alternative activation method. Additionally, platelet activation with thrombin induces aggregation, which creates a PRP gel that can be used in orthopaedic surgery but cannot be injected subcutaneously.[66] A method that activates platelets while limiting aggregation would allow the use of activated PRP as injections in addition to surgery. Garner et al[17,18] have explored the role of electrical stimulation in platelet activation of PRP and whole blood. When combining electrical stimulation with $CaCl_2$, they were able to adjust the strength of the clot, time to clot, and duration of growth factor release.[17] In addition, Irmak et al[24] used a polychromatic light source at 600- to 1200-nm wavelength to activate platelets at regular intervals throughout 28 days. This photostimulation enabled platelets to release significantly higher concentrations of PDGF, bFGF, and TGF-β for the entirety of the 28 days when compared with $CaCl_2$-activated platelets.[24]

Another possible solution for the decline in growth factor concentrations over time is the lyophilization of fresh PRP to create a freeze-dried powder. PRP powder can be stored at room temperature for months while maintaining growth factor concentrations.[4] In addition to its enhanced storage capabilities, PRP powder has been shown to be effective. Hahn et al[21] found that PRP powder increased chondrocyte proliferation and metabolic activity in a dose-dependent manner in vitro. It can be used in an autologous fashion for patients who require multiple applications or could potentially be pooled from healthy donors and be used in an allogenic fashion.[4] Freeze-dried PRP may be able to be standardized and has the potential to eliminate the current challenges of storage and PRP content variability.[4] Overall, decreased efficacy of PRP because of variability of growth factor concentration over time is a limitation of current PRP application methods and is a potential area for more research and innovation.

Platelet Exosomes

In addition to the growth factors, a novel potential explanation for the role of PRP in tissue repair is the ability of exosomes to influence the gene expression of nearby cells (Figure 2). Exosomes are 40- to 100-μm extracellular vesicles that contain the contents of the cell that secretes them, often including transmembrane proteins, lipids, mRNA, and microRNAs.[63] Growth factors, such as bFGF, VEGF, PDGF-BB, and TGF-β1, have also been found in exosomes derived from platelets, indicating exosomes may be a source of the growth factors and therefore may play a significant role in the function of PRP.[65] Iyer et al[26] found that exosomes isolated from PRP accelerated

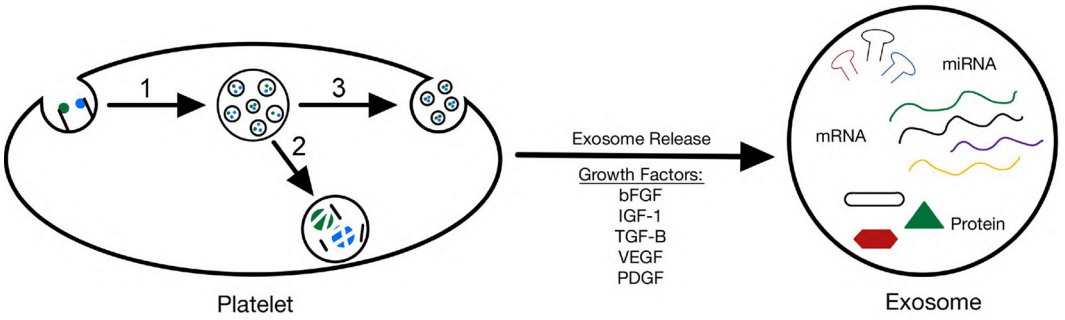

Figure 2. The process of exosome release and exosome contents. (1) Endosomes form from the plasma membrane and are processed into multivesicular endosomes. The multivesicular endosomes can be (2) either degraded by lysosomes or (3) secreted as exosomes. Exosomes can contain miRNA, mRNA, proteins, and growth factors, which can influence nearby cells as a form of intercellular communication and may mediate the effects of platelets in platelet-rich plasma therapy. bFGF, basic fibroblast growth factor; IGF-1, insulin-like growth factor 1; PDGF, platelet-derived growth factor; TGF-B, tissue growth factor beta; VEGF, vascular endothelial growth factor.

TABLE 1
Comparison of Pros and Cons of LR-PRP and LP-PRP[a]

	Pros	Cons
LR-PRP	1. Contains leukocytes	1. May induce pain in injection
	2. Releases more growth factors[32]	2. Elevated catabolic cytokines[60]
	3. Better promotes innervation[43,56]	3. Elevated catabolic proteases[48]
LP-PRP	1. Less pain during use	1. Enhanced fibroblast proliferation[46]
	2. Better promotes angiogenesis[35,66]	2. Less catabolic cytokines and catabolic proteases[60,62]
	3. Anabolic, anti-inflammatory response[72]	3. Role of limitation in tissue regeneration[62]

[a]The difference in the benefits and adverse effects of leukocyte-rich platelet-rich plasma (LR-PRP) and leukocyte-poor platelet-rich plasma (LP-PRP) is potentially accounted for by the difference in leukocyte content and cytokine profile.

muscle recovery after injury in a rat model as evidenced by an increase in centrally nucleated fibers and increased expression of myogenin. In addition to increasing muscle proliferation, exosomes have also been found to prevent glucocorticoid-induced apoptosis in bone in vitro and in vivo by increasing antiapoptotic *Bcl2* gene expression.[61] Furthermore, in an interleukin-1 beta (IL-1β)–induced osteoarthritis model of cartilage in vitro, PRP exosomes increased the migration and proliferation of chondrocytes, decreased apoptosis of chondrocytes, and decreased expression of the proinflammatory cytokine, tumor necrosis factor alpha (TNF-α).[40] A possible method to further elucidate the contents of PRP exosomes is total RNA sequencing. Everaert et al[14] found that the RNA content of extracellular vesicles differs markedly from its biofluid of origin. For example, they found that the exosomes in normal plasma contained 1598 genes, with 70 of those being unique to the extracellular vesicles.[14] RNA profiling of PRP and its exosomes may lead to the identification of biomarkers of clinical significance.[14] The ability of exosomes to induce changes similar to those seen in whole PRP applications suggests that exosomes may play a significant role in the function of PRP, although more research is needed to fully elucidate their role and contents.

Variability of Biological Contents

PRP can differ in the concentration of leukocytes and therefore serve different roles in tissue regeneration (Figure 1). LR-PRP has been shown to release more growth factors, including both anabolic growth factors and proinflammatory cytokines.[32,60] LR-PRP has been shown to improve tendon injury, and LP-PRP has been shown to slow the progression of osteoarthritis.[12,29] Ziegler et al[71] found that LR-PRP contains higher concentrations of IL-1 receptor antagonist (IL-1Ra), an inhibitor of the proinflammatory cytokine IL-1, than LP-PRP, and suggest that, based on this cytokine profile, LR-PRP may be desired for treatment of injuries that require increased angiogenesis and healing, including muscle and tendon. In addition to regenerative properties, other studies indicate no significant difference in improvement of patient pain levels between LR-PRP and LP-PRP therapy.[22] Riboh et al[51] found that LR-PRP and LP-PRP carry a similar risk of adverse effects, indicating that the adverse effects of PRP may not be related to leukocyte concentration. With similar adverse effect profiles and pain reduction levels, the use of LR-PRP versus LP-PRP seems to depend on the target tissue type. Table 1 summarizes the pros and cons for LR-PRP and LP-PRP.

TABLE 2
Summary of the Clinical Applications of PRP in Muscle Healing[a]

	LP-PRP	LR-PRP	PRP Powder[4,19]	Composite/ Engineered PRP[31,33]	Other Factors With PRP + Rehabilitation[47,62]
Pain control[18,39,55,72]	+ +	+ +	+ + +	+ + +	+ + + +
Muscle regeneration[43,46,72]	+ +	+ +	+ +	+ + +	+ + +
Angiogenesis[35,36]	+ + +	+ +	+ +	+ + + +	+ + +
Improving innervation[41,54]	+ +	+ + +	?	+ + +	+ + + +
Adverse effects[35]	–/+	+	+ +	+ +	+ +
Shortened return time back to sports[43]	+ +	+	?	+ +	+ + + +

[a]?, no published data support so far; –/+, little effect; +, some effect; + +, effective; + + +, more effective; + + + +, significantly effective. LP, leukocyte-poor; LR, leukocyte-rich; PRP, platelet-rich plasma.

Beyond leukocyte concentration, other biologic contents can vary based on procedure differences in the preparation of PRP. When comparing PRP products from the same donor using 5 different preparation methods, Magalon et al[41] found that the final PRP products varied significantly in their contents, including differences in the final volumes, platelet concentrations, growth factor concentrations of epidermal growth factor (EGF) and VEGF, white blood cell concentrations, and platelet activation. This variability in the composition of PRP makes it difficult to compare studies that use different preparation methods and could be a potential source of inconsistent results in the literature.

There are also individual patient factors that contribute to PRP variability, such as patient diet and exercise, sex, and the time of day the sample was obtained.[15] Baria et al[6] demonstrated that PRP differs before and after exercise; 4 minutes of high-intensity interval exercise significantly increases the total platelet count and TGF-β concentrations as compared with before exercise. It has also been shown that the efficacy of PRP injections for pain relief may vary with patient sex. A meta-analysis by Zhao et al[69] found that treatment effects were different in subgroups consisting of fewer than 50% women than those consisting of a majority of female patients. They suggested that PRP may not work as well in men as it does in women, although more research is needed in this area.[69] This variability of PRP content, because of varying preparation methods and modifiable and unmodifiable patient variables, may contribute to the inconsistent results regarding the clinical efficacy of PRP application.

BIOLOGICAL APPROACHES FOR SKELETAL MUSCLE INJURY AND REPAIR

Muscle Injuries

Considering the multifaceted nature of tissue healing, PRP has been developed and applied therapeutically to augment the body's natural reaction to injury with the goals of potentially modulating gene expression, inflammation, myogenic response, and apoptosis.[10] To define the context within which PRP can be applied therapeutically, a thorough understanding of the biological mechanisms of skeletal muscle injuries is required.

Skeletal muscle injuries can be grouped into 2 categories: traumatic muscle injuries and contraction-induced muscle injuries.[67] In a study that compared these different injury mechanisms, Warren et al[67] revealed that the mechanism of skeletal muscle injury influences differential gene expression and the extent to which each stage of the healing process occurs. Although this suggests that the natural reaction for tissue healing may depend on the specific mechanism of skeletal muscle injury, the healing process still involves the same events, although the healing results may vary. For instance, traumatic skeletal muscle injuries include strain tears, lacerations, contusions, and ischemic/reperfusion injuries. These various muscle-healing processes can be dependent on the level of injuries; severe injuries result in fibrous scarring at the end of healing.[23,37]

Natural Reaction for Muscle Tissue Healing

The natural process of skeletal muscle repair involves the destruction, regeneration, and remodeling of myofibers and their surrounding environment (Figure 3).[27,28] The destruction phase involves myofiber damage, such as a strain or rupture during sports, usually near the myotendinous junction; necrosis; and an inflammatory reaction in which the macrophages and fibroblasts that direct ECM deposition and necrotic tissue degradation are activated by the actions of myogenic progenitors called muscle stem cells (MuSCs) or satellite cells (SCs).[27,28] Skeletal muscle repair and remodeling is defined by SC proliferation into myoblasts, which can combine to form myotubes that are integrated with injured host myofibers and differentiated into new myofibers.[28] A severe muscle injury can sometimes result in pathological healing, such as a fibrous scar or heterotopic ossification bone formation.[23,36,37] These released inflammation factors and fibrotic factors (eg, TGF-β1) play an essential role in pathological muscle.[38,54] The hematoma forms and provides fibrin and fibronectin in the early stages of scar formation, which make up the granulation tissue that anchors fibroblasts that synthesize ECM components such as tenascin-C, fibronectin, type 1 collagen, and type 3 collagen.[27,28] The requirement of adequate oxygen supply for these processes is met by the vascularization of the injury site.[27,28]

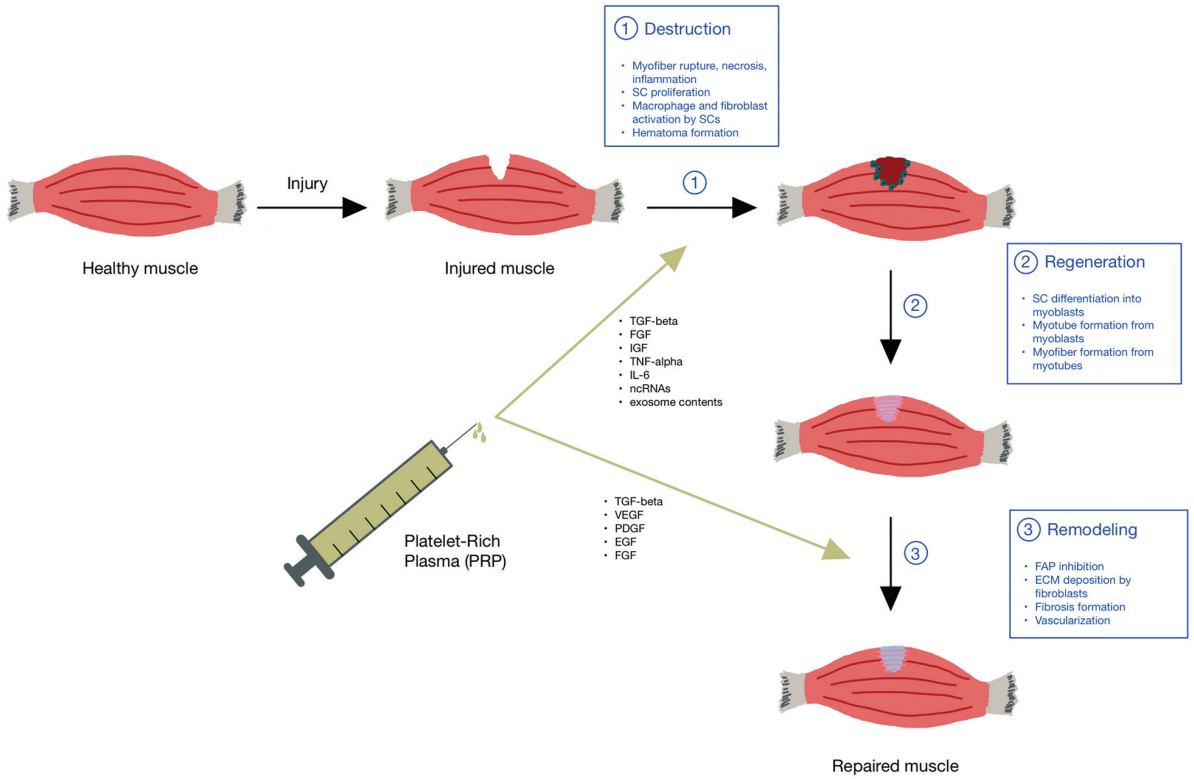

Figure 3. Timeline of muscle healing and a treatment strategy of platelet-rich plasma (PRP) in muscle healing. The schematic illustrates the hallmarks of each phase of muscle tissue repair and indications of where unmodified PRP contents potentially augment the process. Of note, unmodified PRP has not been shown to play a role in satellite cell (SC) and myoblast differentiation in muscle tissue regeneration. ECM, extracellular matrix; EGF, epidermal growth factor; FAP, fibroadipogenic progenitor cell; FGF, fibroblast growth factor; IGF-1, insulin-like growth factor 1; IL-6, interleukin-6; PDGF, platelet-derived growth factor; CTGF, connective tissue growth factor; TGF-beta, tissue growth factor beta; TNF-alpha, tumor necrosis factor alpha; VEGF, vascular endothelial growth factor.

Additionally, a variety of growth factors and cytokines such as FGF, IGF, TGF-β, TNF-α, interleukin-6 (IL-6), and several others are thought to be involved in this natural reaction for tissue healing in many ways, including activating SC proliferation.[28,54] The application of PRP that contains various growth factors can be involved in the stimulation of the resident MuSCs and SCs at wound sites. Sass et al[54] highlight the cellular interdependencies that integrate the roles of these factors. For instance, macrophages and SCs stimulate each other through the release of different factors to degrade necrotic tissue and proliferate as myofiber precursors, respectively. Also, fibroblasts, MuSCs, and/or SCs stimulate each other to coordinate ECM deposition and muscle regeneration.[54] Zhao et al[70] report that several noncoding RNA molecules also play key roles in regulating the quiescence, activation, proliferation, and differentiation of MuSCs, suggesting the complexity of healing injured skeletal muscles.

Pathological Healing and Fibrosis Formation

Despite the natural reaction to skeletal muscle injury, fibrosis generation is a pathological process that negatively affects tissue healing and function. Fibrosis production is mediated by the action of specialized cells that produce an accumulation of ECM components in traumatically injured muscles. Namely, muscle injury elicits the excessive deposition of ECM through the actions of growth factors such as TGF-β1, connective tissue growth factor, PDGF, VEGF, EGF, FGF, and myostatin.[42] In conjunction with the proinflammatory cytokines, TNF-α and IL-6, these molecules promote cellular differentiation, proliferation, and survival while inducing ECM protein production and inhibiting ECM degradation, leading to the accumulation of ECM material that characterizes fibrosis.[42] It is worth noting that the extent to which fibrosis occurs is greater in the context of immobilization of the injury site and surrounding structures.[28]

The TGF-β family of growth factors is a particularly important mediator of skeletal muscle fibrosis because of its various roles in the process of formation. Downstream signaling by TGF-β can elicit differentiation of multiple cell types into myofibroblasts that produce collagen and other ECM material.[25] Studies have also shown that fibroblast proliferation and ECM synthesis are promoted by TGF-β–mediated modification of transcription factor expression, such as Sharp-1 and scleraxis.[25] Additionally,

TGF-β–mediated suppression of fibroadipogenic progenitor cell (FAP) apoptosis sustains fibrosis formation in excess of the tissue healing response that these cells normally facilitate.[25]

MuSCs are myogenic progenitors that contribute to myofiber fusion and muscle regeneration and ECM production in an optimal environment created by FAPs.[16] The inhibition of FAP apoptosis can impair proper MuSC myogenic differentiation and lead to excessive ECM deposition by MuSCs. In conjunction with this, FAPs secrete IL-6, a cytokine that is heavily implicated in skeletal muscle fibrosis.[16] According to several studies, sustained IL-6 secretion is thought to promote collagen expression, enhance TGF-β signaling, and suppress the promyogenic functions of IGF-1,[16] demonstrating the complexity of fibrosis formation and concomitant inhibition of proper skeletal muscle tissue healing. Considering the negative implications of fibrosis on tissue healing, function, mobility, and risk for reinjury, it is critical to be able to mitigate pathological fibrosis in the process of proper tissue healing. However, the application of PRP alone or combined with antifibrosis agents can eliminate fibrous scars in those injured skeletal muscles,[2,34] although there are controverted reports that suggest that the application of PRP has no role in preventing fibrosis in injured muscles.[64]

PRP CLINICAL APPLICATIONS IN MUSCLE HEALING

Use for Pain Control/Relief

There has been increasing interest in the clinical effectiveness of PRP injections on different muscular injuries as presented in Table 2. One possible benefit is a reduction in pain. Pain from muscle injury is initiated from the proinflammatory mediators during muscle healing, such as TNF-α, IL-1, IL-6, and substance P.[39,56] PRP is able to regulate those inflammatory mediators and thus reduce pain.[19,55] The mechanism behind the beneficial effects of PRP is thought to start by expediting the inflammation phase.[8] Platelet contents are released, attracting leukocytes, neutrophils, macrophages, and fibroblasts to the injury site to clear debris and induce progression to the subsequent phase of healing. Clinically, this would result in decreased pain, allowing athletes to recover quicker and return to play faster.

Previous investigations into the effectiveness of PRP in pain reduction have shown varying results. PRP use in lateral epicondylitis has shown significant pain reduction, while rotator cuff repairs did not show significant pain reduction.[11] Some of the studies have also reported decreased swelling and pain clinically.[1,7,9] A recent meta-analysis by Grassi et al[20] investigating the effectiveness of PRP for acute muscle injuries included various studies evaluating pain reduction. They found that 3 out of 5 studies showed significant pain reduction, especially in the first 3 weeks. The authors concluded that there was no difference in pain reduction at final follow-up and a possible increased range of motion in the first 4 weeks. None of their studies found a difference in imaging after PRP therapy but reported decreased time to return to play. Sheth et al[57] performed a meta-analysis on randomized controlled trials performed from 2013 to 2016 and concluded that PRP injections significantly reduced the time to return to sports by about 1 week without increasing the risk of reinjury. This is a clinically significant time for high-level athletes, who could participate in more competitions in that time frame. Many studies have reported a change in the time required to return to play without any change in the muscular structure/regeneration compared with the control.[55] The reduction in pain, allowing earlier range of motion and recovery exercises, is the most likely cause for the earlier return to play.

There are many limitations to the previous studies on the effect of PRP on muscle injury as it relates to pain management. There continue to be nonstandard protocols for the preparation of PRP. Despite the different stages of muscle healing (Figure 3), there has been no emphasis on the timing of administration of the PRP injection, to our knowledge. Only 2 studies reported the timing of initial injection; Rossi et al[52] reported a mean time to injection after onset injury of 2.3 days, and A Hamid et al[2] reported a mean injection time of 4.6 days. With all these differences in study design, it is difficult to compare different trials with each other.

Accelerating Muscle Regeneration

Muscle regeneration and prevention of fibrosis scarring is the ultimate goal of treatment of acute muscle injury. Grassi et al[20] found that none of the recent randomized controlled trials showed a significant difference in the structural change of the muscle through ultrasound or magnetic resonance imaging after PRP injection. Currently, PRP has not been shown to be clinically effective in regenerating muscle, but the answer could be in the basic science. PRP without modification has been shown to increase myoblast proliferation, but not differentiation.[35] Differentiation is required to produce new muscle tissue to facilitate the repair. Some growth factors found within PRP, including myostatin and TGF-β, inhibit this process.[5,35] This might point to a contraindication to PRP injection during the remodeling phase because of its ability to induce muscle fibrosis by increasing the concentration of TGF-β. Two studies have shown that PRP with a TGF-β inhibitor or antibody was able to induce differentiation in vitro.[30,34] Another study found that PRP with an extra round of centrifugation to remove platelets was also able to induce myoblast differentiation.[10] With TGF-β inactivated, fibrosis and scarring of the muscle will be decreased.[2,34] Although these ideas have not been proven in vivo, they show the potential of selecting for or inhibiting certain signaling molecules within PRP. Faster muscle regeneration and prevention of fibrosis through a series of engineered PRP injections could pave the path toward faster recovery and return of function. Clinical applications of engineered PRP injections could be applied to any acute muscle injury. In addition, some other factors involved with PRP injection, such as rehabilitation, significantly improved the return time back to sports of injured

athletes.[1,57] These interesting strategies and potential mechanisms are under investigation.

Improving Functional Repair via Vascularization and Innervation

In order for PRP to be an effective treatment, the regenerated muscle must be both histologically healthy and functional after recovery. Therefore, the ability of PRP injections to induce active repair after acute muscle injury relies on its ability to promote the regeneration of vessels and nerves. Repair of any tissue requires adequate nutrients, inflammatory cells, and growth factors, which are provided by the blood. In addition, skeletal muscle requires innervation to prevent atrophy and degeneration. One potential benefit of PRP injection is the increased concentration of VEGF, the main growth factor responsible for angiogenesis. Increased vascularization of damaged tissue leads to improved repair. Previous research on the effect of PRP injections of tendons has shown mixed results regarding vascularization,[43] but this has not been investigated thoroughly in muscle tissue. There has also been interest in the angiogenic properties of platelet-derived fractions for use in tissue engineering.[44] In addition to proper blood supply, axon progression to the new myofibrils is necessary to regain function of the skeletal muscle. This can be hindered by granulation tissue and fibrosis.[53] There has been interest in the effect of PRP in nerve regeneration, specifically in the realm of nerve grafting in plastic surgery.[59] Research found that topical application of PRP increased the number of myelinated axons and could play a role in nerve regeneration.[68] The role of PRP injections in nerve regeneration can be 2-fold: removing barriers of axon progression and increasing the number of axons progressing toward the site of muscle injury. Adequate innervation of the skeletal muscle after injury is necessary to regain functional repair and could be an additional clinical application of PRP injections.

CONCLUSION

The autologous blood product, PRP, is a focus point of recent research in regenerative medicine and has been widely used in clinical application. The medical field has adopted this because its autologous use requires minimal resources, and it is a relatively simple procedure to perform with minimal complications. There are many studies that have investigated the mechanisms behind PRP, which suggest growth factors and active proteins could have a major effect in repairing injured tissues as well as preventing pain. Despite containing growth factors that stimulate pain (eg, ILs) and fibrosis (eg, TGF-β) in the target tissues, the functional promotion of tissue regeneration of PRP has been evaluated in many studies. However, PRP has a limited role in muscle fibrosis prevention that is a major controversial discussion in the application of muscle healing.[43,64,72] In our review, we discussed some of the more recent studies in the basic science behind PRP as well as the potential application in orthopaedics and sports medicine. The application of PRP in skeletal muscle injury and repair has been well investigated, specifically its role in stimulating muscle regeneration and controlling pain, but it requires further research and possible implementation of modified PRP products to take advantage of the basic science principles. Furthermore, storing fresh human PRP for autologous applications is still a challenge. New investigations into lyophilization and creation of PRP powders[4] may be of some benefit, but further work is needed in this area.

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